Comments on the National Toxicology Program Draft Report on Carcinogens Substance Profile for Formaldehyde

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1.0 Introduction

The National Toxicology Program (NTP) staff has prepared two documents for public review and comment in support of the listing of formaldehyde in the 12th Report on Carcinogens. The first is the Background Document (NTP 2010a) on the toxicity of formaldehyde, which contains summaries of the epidemiological, toxicological and mechanistic data available at the time for its initial draft in September, 2009. This draft document was finalized in January, 2010 and it is important to note that this document, which provides a summary of the underlying science for reaching any conclusions regarding the potential for formaldehyde to be causally associated with leukemia and other lymphohematopoietic cancers, was finalized prior to the end the public comment period. It was also finalized prior to the completion of the public comment period on the External Expert Panel report, which further summarized the data reported in the Background document and makes a recommendation regarding the listing of formaldehyde. The second document prepared by the NTP staff is the Draft Substance Profile (NTP 2010b), which provides the NTP's preliminary recommendation on the listing status of formaldehyde in the Report on Carcinogens and a further summary of the scientific information from the Background Document (NTP 2010a) and the Expert Panel report.

ENVIRON International Corporation, on behalf of Hexion Specialty Chemicals, Inc., has previously provided comments on the Background Document and Expert Peer Review Panel Report and is now providing comments on the Draft Substance Profile (NTP 2010b). In our initial comments on the Background Document (2010a), we stated that there was a lack of clarity in the summarizations of the data on which of the reported epidemiological studies were statistically significant and how the lack of statistical significance affects interpretation of these results. Unfortunately, this lack of consideration of the strength of the data has been carried forward into the Draft Substance Profile (NTP 2010b), resulting in conclusions that are not supported by the epidemiological data for formaldehyde with regard to the potential association of formaldehyde exposure and myeloid leukemia or other lymphohematopoietic cancers. NTP has acknowledged in the Draft Substance Profile (NTP 2010b) that the mechanisms by which formaldehyde causes myeloid leukemia in humans are not known. The strength of the epidemiological evidence that formaldehyde causes myeloid leukemia is exaggerated by NTP, and the NTP has not adduced the evidence that would be needed to support the biological plausibility for such an effect for a compound, such as formaldehyde, that is endogenously present. This results in a Draft Substance Profile (2010b) that contains broad generalized statements that do not reflect the results of the cited studies. Nor does it present a scientifically balanced, integrated interpretation of the published data. Because of this lack of balance, a number of the statements and conclusions in the NTP (2010) document require additional critical review.

2.0 Comments on Section *Cancer Studies in Humans – Myeloid Leukemia*

The epidemiological studies that have evaluated the potential association between formaldehyde exposure and lymphohematopoietic cancers and/or leukemia are studies conducted by the National Cancer Institute (NCI) (both industrial and professional workers) (Beane Freeman et al. 2009; Hauptmann et al. 2009), the National Institute of Occupational Health and Safety (NIOSH) (industrial – garment workers) (Pinkerton et al. 2004), and a study in the United Kingdom (UK) (industrial workers) (Coggon et al. 2003). The Substance Profile has drawn the following conclusions that are not supported by the epidemiological data.

- **The Draft Substance Profile (NTP 2010b) states: "An association between excess mortality from leukemia or combined lymphohematopoietic cancer has been**

reported in numerous cohort studies, including all of the studies of professional groups and some of the industrial cohorts"

With regard to a causal association between formaldehyde exposure and all lymphohematopoietic cancers, one of the four studies (Coggin et al. 2003) relied upon by the NTP staff in reaching their conclusion with regard to lymphohematopoietic cancers did not evaluate this association.

- Exposure to formaldehyde was **not associated** with a significant increase in all lymphohematopoietic cancers in the studies conducted by Pinkerton et al. (2004) in garment workers, or Hauptmann et al. (2009) in funeral directors and embalmers.
    - In the Hauptmann et al. (2009) study, there was neither a statistically significant association with formaldehyde exposure nor a statistically significant trend, when categorized by number of embalmings, average, cumulative, or peak exposure (up to greater than 9.3 ppm), when compared to the referent group of persons with less than 500 lifetime embalmings. A significant trend was noted when the cohort was categorized by duration of working in jobs with embalming; however, the association was not significant for other exposure categories. All confidence intervals included 1.0.

- Although exposure to formaldehyde was associated with a statistically significant increase for all lymphohematopoietic cancers combined in studies in the NCI cohort reported by Hauptmann et al. (2003) and Beane Freeman et al. (2009), this was the case only in workers in the highest "peak" exposure category (greater than 4 ppm) and not when other dose-metrics were considered, such as average or cumulative exposure or duration of exposure. No association with average exposure was noted when the Hauptmann et al. (2003) results were re-evaluated by Marsh and Youk (2004) and when updated with ten additional years of follow-up by Beane Freeman et al. (2009). More than 1000 deaths among cohort members were not included in the previous investigation (Hauptman *et al.* 2003) with proportionally more deaths missed among the unexposed and non-exposed groups (Marsh and Youk 2004). Fundamental questions remain:

    - ***A fundamental question is the biological basis for combining all of these diseases into one category.*** Acute diseases are different from chronic diseases, and lymphocytic cancers arise in different cells than myelogenous cancers (lymphoid cells versus myeloid cells, respectively). Leukemia is different from lymphoma. Leukemia arises in both myeloid cells and lymphoid cells, while lymphoma arises only from lymphoid cells. These diseases have different etiologies. The processes that lead to these diseases are distinct, and each disease has different risk factors and therefore, should not be combined for evaluation.

    - ***The characterization of "peak" exposure is uncertain.*** According to Hauptmann et al. (2003), no measures of peak exposure were available for this cohort. Peak exposures for the entire NCI cohort were estimated by an industrial hygienist based on job and task descriptions compared to a Time-Weighted 8-hour Average.
        - As discussed by Marsh and Youk (2004),

        "The authors essentially treated peak exposure (more accurately, highest peak exposure) as a monotonically increasing, time-dependent variable. That is, as subjects moved through age-group- and time period-specific person-time counts, persons and person-years were also allocated to the highest peak exposure category experienced at that time. Unless the subject subsequently works in a job associated

with a higher peak exposure, all subsequent person-years are allocated to the highest peak category attained previously. Consequently, many different patterns of peak exposure are possible that would result in a given leukemia death being assigned to the same highest peak category."

- As also discussed by Marsh and Youk (2004), the rationale for the peak exposure category ranges, i.e., >.1 to 1.9, 2.0 to 3.9, and equal to or greater than 4.0 ppm was not provided. Other exposure categories could have been used, possibly with different results (see discussion under myeloid leukemia).

- ***Comparison of the higher exposed groups to the lower exposed group within the cohort rather than to external local or US-based mortality rates also introduces uncertainty*** (see discussion under myeloid leukemia).

- **The Draft Substance Profile (NTP 2010b) states: "Among studies that evaluated subtypes of lymphohematopoietic cancer, the strongest associated were observed for myeloid leukemia."**

  - In the Beane Freeman et al. (2009) study, the incidence of neither leukemia nor myeloid leukemia was statistically significantly increased by <u>any</u> of the dose metrics evaluated, i.e., peak exposure, average exposure concentration, cumulative exposure concentration, and duration of exposure, nor was the trend across exposure categories significantly increased for any of these dose-metrics.

  - In the reanalyses of Hauptmann et al (2003) original data, Beane Freeman et al. (2009), and Marsh and Youk (2004), found no significant increase in either endpoint in any alternative exposure categories: average intensity, cumulative exposure, duration of exposure, duration of time worked in the highest peak category, or time since first highest peak exposure. Marsh and Youk (2004) did not find a statistically significant increase in leukemia or myeloid leukemia when compared to external mortality rates (SMRs), and suggested that the positive findings in Hauptmann *et al.* (2003) were due to statistically significant deficits in deaths in the baseline group, an observation that was confirmed by Beane Freeman et al (2009). More than 1000 deaths among cohort members were missed in the previous investigation (Hauptman *et al.* 2003), with proportionally more deaths missed among the unexposed group.

  - Pinkerton et al. (2004) did not find a statistically significant increase in either leukemia or myeloid leukemia when the entire cohort was evaluated. Although a significant increase in myeloid leukemia, but not all leukemia, was reported when the cohort was categorized by time since first exposure for workers with 20+ years of exposure, this association was no longer significant when acute myeloid leukemia was evaluated separately. Neither all leukemia, myeloid leukemia, nor acute myeloid leukemia were significantly increased when evaluated by either the duration of exposure or by the year of first exposure.

  - Hauptmann et al. (2009) reported findings similar to those of Pinkerton et al. (2004).

    - Mortality from myeloid leukemia was significantly increased in embalming workers employed for greater than 34 years duration but was no longer significant when acute myeloid leukemia was considered separately
    - Similarly, the authors reported a significant increase in myeloid leukemia among those workers who either performed more than 3,000 embalmings, or who had a cumulative exposure of more than 9200 ppm-hours. These elevations were modest,

with a lower bound on the confidence interval of 1 in both cases. Based on the typical definition of statistical significance, the second two categories would not be considered statistically significant because the confidence intervals do not exclude 1.0.

- Further, mortality from acute myeloid leukemia was not significantly increased in any of the exposure categories considered.

- Numerous methodological limitations have been noted for Hauptmann et al. (2009), including:

  - Although some effort was made to assure comparability between the cases and controls overall, important differences are apparent, specifically between myeloid leukemia cases and the control group. For example, myeloid leukemia cases were 50% more likely than controls to have begun employment in the funeral industry prior to 1942. This suggests that the myeloid leukemia cases were generated by an older and earlier source population than is represented by the controls, and may explain why this group performed more embalmings. This and other differences between cases and controls, including earlier year of death, all white race, longer time employed and age first employed, appear not to have been adequately considered by the authors.

  - As reported by the authors, the standard statistical analyses initially reported are unreliable due to the fact that there was only one unexposed myeloid leukemia case, leading to very large and unstable confidence intervals. Adjusting the lowest exposure group to include cases and controls performing fewer than 500 estimated embalmings resulted in drastically reduced Odds Ratios (ORs) that the authors consider more realistic. However, no clear rationale is provided for using less than 500 embalmings as an "unexposed" group, and most exposed groups produce roughly similar ORs regardless of the exposure category.

  - Contrary to most results presented, myeloid leukemia cases and controls had nearly identical mean estimated average formaldehyde exposure, TWA-8 hour exposure, and peak formaldehyde exposure. As expected, estimated number of embalmings and the correlated cumulative exposure were slightly higher for cases – likely due to their earlier first employment, younger age at hire and older age at death, leading to longer average employment in the industry.

- **Coggon et al. (2003)** is a study of more than 14,000 workers employed in factories where formaldehyde was manufactured or produced. No significant increase in leukemia (type not specified) was found even when the cohort was stratified by duration of exposure or when the cohort was limited to only those workers considered to be in the high-exposure group. In fact, there was a deficit in leukemia in the exposure group >2 ppm. Formaldehyde exposures were generally higher for these workers than those included in the NCI study, and a higher percentage of workers were considered to be in the "high" exposure category compared to the NCI cohort. Although NTP (2010b) acknowledged that this study did not show an association with leukemia, the NTP (2010b) provided no discussion/justification why these results did not influence their conclusions.

- **NTP (2010b) did not consider the results of the published meta-analyses in reaching their decision to classify formaldehyde as a "known" human carcinogen.**

As stated, NTP (2010b) apparently relied on just four studies in reaching their conclusion regarding the evidence for causation of myeloid leukemia. There was no consideration of 12 additional cohort studies with leukemia findings among formaldehyde-exposed workers. When a number of studies have been conducted that may have dissimilar designs and different results, meta-analysis is an accepted statistical procedure to aid in reaching a strength-of-evidence conclusion based on epidemiological data. The most recent and comprehensive meta-analysis, conducted by Bachand *et al.* (2010), evaluated 18 studies in which an association was investigated between formaldehyde exposure and leukemia, in particular myeloid leukemia, and concluded that formaldehyde is not causally associated either with leukemia or myeloid leukemia (Figure 1).

Three other meta-analyses have been conducted since 2004 (Bosetti *et al.* 2008; Collins and Lineker 2004; Zhang *et al.* 2009). These studies did not consider the most recent data from the NCI cohort published by Beane Freeman *et al.* (2009). These studies did not adequately investigate potential sources of heterogeneity other than the effect of job type, and they did not include sensitivity analyses that could have been conducted with the available data, and included proportional mortality rates (PMRs) which are not useful in determining causal associations (Bachand *et al.* 2010). The NTP discounted the results of the Bachand et al. (2010) study because it did not include studies that relied on PMRs. Bachand et al. (2010) provided clear justification for the exclusion of PMR studies. Bachand (2010) excluded results from PMR studies because such studies are considered limited for evaluating causation and may best be viewed as a hypothesis-generating exercise. As noted by Checkoway (2004):

> PMR studies have the attractive feature of providing results relatively quickly. However, the validity of a PMR study depends on whether the deaths included are generally representative of all deaths that would be identified if follow-up of the full cohort had been conducted. For example, if deaths from a particular cause were recorded preferentially because they were compensable or of particular prior concern, then the PMR for that cause would probably appear to be elevated, even if the actual rate of disease in the cohort were not excessive.

The subjectivity that PMR studies introduce into a meta-analysis (risk estimates derived from groups that may not be representative of the US population as a whole) could preferentially bias the meta-risk estimates toward an association, even if, as in this case, there is a large body of evidence to the contrary.

In summary, the NTP (2010b) interpretation of findings for leukemia did not consider the totality of results available, and simply ignored the lack of statistical significance of the observations. The strength of the evidence for a causal association between formaldehyde exposure and myeloid leukemia is lacking.

3.0 Comments on Section *Studies on Mechanisms of Carcinogenesis – Myeloid Leukemia*

Since the first epidemiology studies suggesting a possible association between formaldehyde exposure and leukemia were published, a major area of debate has been how mechanistically formaldehyde could cause a disease that develops distant from the point of contact. Formaldehyde is rapidly metabolized and highly reactive and, because it is an endogenous compound, a detectable change in the natural background levels would need to occur in order to result in the potential for adverse effects. Although, the Draft Substance Profile (NTP 2010) cites hypotheses proposed by Zhang *et al.* (2009) regarding the theoretical development of leukemia following inhalation of formaldehyde, there is no documented evidence to support the validity of these hypotheses. In fact, Zhang *et al.* (2009, 2010) note that their hypotheses related to mechanisms of leukemia clearly require additional testing. The existing

mechanistic data for formaldehyde provide no evidence that exogenous formaldehyde will be transported from the point of contact to distant sites, but do provide evidence that formaldehyde does not affect the relevant target cells (bone marrow) for leukemia.

- **Evidence for systemic distribution of formaldehyde, an endogenously present compound, does not provide evidence of adverse effects, specifically myeloid leukemia.**

Pala et al. (2008) is the only study cited as providing evidence of systemic distribution of formaldehyde-albumin adducts in laboratory workers following exposure to high levels of formaldehyde. Pala *et al.* (2008) evaluated potential relationships between formaldehyde exposure in the workplace and biomarkers of exposure (formaldehyde human serum albumin conjugate) and biomarkers of effect (chromosomal aberrations, sister chromatid exchanges, and micronucleated cells) in 36 workers exposed to formaldehyde in a cancer research facility. A statistically significant relationship between formaldehyde exposure, measured with personal samplers, and biomarkers of exposure was noted. However, the authors reported **no statistically significant relationship between formaldehyde exposure and biomarkers of effect (i.e., chromosomal aberrations, sister chromatid exchanges, and micronucleated cells). Furthermore, the methods used do not differentiate between formaldehyde of endogenous and exogenous origin.**

- **The relevance of effects, such as micronuclei and chromosomal aberrations, in circulating lymphocytes to the mechanism of myeloid leukemia has not been demonstrated.**

While several studies are cited in the Draft Substance Profile (NTP 2010b) as demonstrating cytogenetic changes (i.e., micronuclei, chromosomal aberrations) in peripheral lymphocytes, just as many studies provide no evidence of these effects in humans (Bauchinger and Schmid 1985; Fleig et al. 1982; Thomson et al. 1984; Vasudeva and Anand 1996; Ying et al. 1997, 1999; NTP 2010a). In addition, selected studies, while positive for exposed versus control subjects did not demonstrate any differences with increasing exposure to formaldehyde (Shaham et al. 2003). Because it has been demonstrated that exogenous formaldehyde does not result in an increase in formaldehyde concentrations in the blood (Heck et al. 1985), genetic differences in the study populations or difference due to diet or exposure to other compounds (e.g., from embalming fluid) cannot be excluded from consideration as possible causes of the effects reported (Heck and Casanova 2004).

- **Toxicity noted at distal sites is not related either to the potential for formaldehyde to cause myeloid leukemia or a mechanism by which formaldehyde may cause myeloid leukemia.**

While toxicity has been noted at distal sites in mice and rats following inhalation exposure to formaldehyde (NTP 2010a, b), the exposure in these studies was to high concentrations of formaldehyde ranging from 5 to 20 ppm (Cikmaz et al. 2010; Aslan et al. 2006; Sarsilmaz et al. 2007; Ozen et al. 2005). It is critical to note that, in the chronic studies conducted to evaluate the potential carcinogenicity of formaldehyde in rats, mice, hamsters and monkeys, the only exposure related tumors were reported in the nasal cavity (primarily squamous-cell carcinoma) (NTP 2010a). The exposure concentrations in these carcinogenicity studies were as high as 15 ppm, with no other exposure-related tumors reported. In addition, no adverse hematological effects have been noted in subchronic or chronic studies in experimental animals (NTP 2010a; Appleman *et al.* 1988; Dean *et al.* 1984; Johannsen *et al.* 1986; Kamata *et al.* 1997; Kearns *et al.* 1983; Til *et al.* 1988, 1989; Tobe *et al.* 1989; Vargova *et al.* 1993;

Woutersen *et al.* 1987). Therefore, because no carcinogenicity at sites distal from the point of entry were observed following exposure to high concentrations (>5 ppm) of formaldehyde, toxicity at distal sites provides no evidence of the potential carcinogenicity of formaldehyde and the relationship of toxicity in remote tissues to the potential mechanism of myeloid leukemia is unclear.

- **Neither formaldehyde, nor methanediol resulting from inhaled exogenous formaldehyde, have been detected at distal sites such as bone marrow, white blood cells, lung, spleen, liver or thymus.**

The Draft Substance Profile (NTP 2010b) describes the formation of methanediol, a hydrate formed when formaldehyde reacts with water, as a mechanism by which a reactive chemical, such as formaldehyde, can be distributed and undergo metabolism throughout the body. However, when absorbed after inhalation or ingestion, very little formaldehyde reaches the systemic circulation because it is rapidly metabolized at the site of absorption to formate, which is excreted in the urine or oxidized to carbon dioxide and exhaled (NTP 20010a). Recent work by Swenberg and colleagues (Lu *et al.* 2010) evaluated the formation of DNA adducts resulting from endogenous formaldehyde, versus those formed from exogenous formaldehyde in nasal tissues, liver, lung, thymus, spleen and bone marrow in rats following inhalation exposure to 10 ppm [$^{13}CD_2$]-formaldehyde for 1 or 5 days. In addition, the method used monitored transitions that would occur if there was any hydrogen-deuterium exchange, enabling the evaluation of potential transfer of methanediol from the portal of entry. No exogenous formaldehyde-induced DNA adducts were detected in any distant tissue even though up to 5 times greater amounts of DNA were used, while the DNA adducts from endogenous formaldehyde were present in all tissues examined in similar amounts. In fact, endogenous adducts were present at 2.5-3-fold greater amounts than exogenous adducts in the respiratory nasal epithelium. Lu *et al.* (2010) concluded that the results do "not support the biological plausibility for the causation of leukemia". Other studies (NTP 2010a; Wang et al. 2009) of formaldehyde-DNA adducts in the blood did not distinguish between the contribution from exogenous versus endogenous formaldehyde.

- **The biological relevance of the chromosomal changes monosomy 7 or trisomy 8 demonstrated in cell cultures *in vitro* are unclear.**

In ***individuals already presenting*** with acute myeloid leukemia (AML) or myelodysplasia (MDS), trisomy 7 is present in 16-17% of cytogenetically abnormal cases of AML and MDS (Paulsson and Johansson 2007), and monosomy 7 occurs in 10-14% of cases (Johnson and Cotter 1997). In 200 patients ***with AML***, only a limited number of patients had monosomy 7 (2.2%) and trisomy 8 (5.6%) (Ahmad *et al.* 2008). In addition, in 122 ***AML patients*** in China, monosomy 7 was not reported, and only approximately 3% had trisomy 8 (Zheng *et al.* 2007). For chronic myeloid leukemia (CML), translocation of chromosomes 9 and 22 of the Philadelphia chromosome is the chromosomal change most commonly found (Bonassi *et al.* 2008).

Zhang *et al.* (2010) reported an increase in the incidence of monosomy 7 and trisomy 8 in colonies of ***cultured granulocyte-macrophage colony-forming units*** taken from 10 "highly" (undefined) exposed ***healthy*** workers in two factories that produced formaldehyde-melamine resins. It is very important to recognize that these changes were not identified in the individuals and that they were formed during *in vitro* cultivation. It is also important to note that these changes were also observed in the "control" cultures; therefore, the biological relevance of these *in vitro* observations is unclear.

Zhang et al. (2010) indicate in the text that ***unadjusted summary measures*** are presented for all endpoints; therefore, the results presented in tables and figures have not been adjusted for relevant covariates nor were they related to the amount of formaldehyde to which the workers were exposed.

While the small cohort was said to have been matched by age, only 10 exposed and 12 nonexposed individuals were included in Zhang et al. (2010) and the results reported were not adjusted for age of these subject. Because these chromosomal changes reported by Zhang *et al.* (2010) are not commonly observed in myeloid leukemia patients, and because these changes were only detected after *in vitro* culture of stem cells, additional studies are needed to demonstrate that these changes are associated with formaldehyde exposure. It is our understanding that Zhang and colleagues have secured the funding to conduct their investigation with a larger number of workers.

- **Presence and/or frequency of cytogenetic damage, i.e., micronuclei and chromosomal aberrations in the peripheral blood, are not a validated marker of myeloid leukemia.**

The Draft Substance Profile (NTP 2010b) cites Bonassi et al. (2008) and Murgia et al. (2008) as providing evidence that high levels of chromosomal aberrations and micronuclei are associated with increased cancer risks in otherwise healthy individuals. In Bonassi *et al.* (2008), which includes the genetic screening in 22,358 ***cancer-free*** individuals with follow-up for an average of 10 years, the only cancer site significantly associated with the frequency of chromosomal aberrations was stomach cancer (Bonassi *et al.* 2008). In particular, no significant association between cancers of the lymphohematopoietic system and the frequency of chromosomal aberrations was reported by Bonassi *et al.* (2008). In Murgia et al. (2008), no incidences of leukemia were reported; therefore, no conclusions on the association between the presence of cytogenetic damage and leukemia may be drawn from this study.

- **The proposed indirect mechanisms by which formaldehyde could cause leukemia require additional studies to determine whether they are biologically plausible.**

The Draft Substance Profile (2010b) provides two hypothetical mechanisms, reported by Zhang et al. (2009), by which formaldehyde might cause leukemia in the absence of any direct contact with bone marrow. The first suggests that formaldehyde might damage stem cells circulating in the blood, which might travel to the bone marrow and become initiated leukemia cells. However, formaldehyde normally is present in human blood at concentrations of 2 to 3 μg/g, and it has been demonstrated that these levels do not increase after ingestion or inhalation of formaldehyde from exogenous sources (NTP 2010a; Heck et al. 1985). Therefore, it is not biologically plausible that exogenous exposure to formaldehyde, which is ***normally present*** in the blood, would result in an adverse effect.

The second proposed mechanism suggests damage to stem cells that reside in the nasal turbinates or olfactory mucosa, with the implication that these damaged cells could indirectly impact bone marrow. A study by Murrell et al. (2005) is cited in the Draft Substance Profile (NTP 2010b) to demonstrate that olfactory epithelial cells obtained from nasal passages may repopulate hematopoietic tissue of irradiated rats. Radiation exposure can result in destruction of both the blood-forming bone marrow cells, as well as apoptosis of the cycling hematopoietic cells (Kopp et al. 2005). Injected hematopoietic stem cells (HPCs) or progenitors cells (HPCs) have been used successfully for bone marrow transplantation by intravenous infusion. However, this process requires the injection of selected antigens, adhesion molecules and other factors at high levels to produce efficient "homing" or transfer of the circulating HSCs/HPCs back into the bone marrow niche (Kopp et al. 2005). The Murrell et al. (2005) study was an experiment to determine if progenitor cells from an alternate tissue can serve to help repopulate the bone marrow following destruction of the normal progenitor cells in the bone marrow and circulating blood system that typically serve that function. These cells from another tissue (olfactory) were injected intravenously into the irradiated animals. This study does not provide evidence that these olfactory cells could be transported via normal biological mechanisms from the olfactory tissue to the bone marrow and there repopulate normally functioning bone marrow. Rather, at this time, studies are needed to demonstrate that this type of transport of cells could occur in a normal biological setting.

Although, the Draft Substance Profile (NTP 2010b) cites hypotheses proposed by Zhang *et al.* (2010) regarding the theoretical development of leukemia following inhalation of formaldehyde, there is no documented evidence to support the applicability of these hypotheses. In fact, Zhang *et al.* (2010) note that their hypotheses related to mechanisms of leukemia clearly require additional testing. The existing mechanistic data for formaldehyde, which consider the toxicokinetics and genotoxicity of a compound, provide no evidence that exogenous formaldehyde will be transported from the point of contact to distant sites, however, these data do provide evidence that formaldehyde **does not** affect the relevant target cells (bone marrow) for leukemia (Lu et al. 2010).

In summary, insufficient evidence is available to support a potential mechanism for the development of myeloid leukemia following inhalation exposure to formaldehyde.

4.0 Conclusions/Recommendations

In evaluating the available epidemiological, toxicological, and mechanistic data for formaldehyde, the strength of the evidence does not justify classifying formaldehyde as a "known" human carcinogen based on myeloid leukemia. Overall, the epidemiological data reviewed by NTP (2010b) do not show a causal relationship between formaldehyde exposure and myeloid leukemia. The mechanistic data do not support significant transfer to or effects of exogenous formaldehyde in tissues distant from the point of contact, and there remain significant uncertainties regarding a mechanism of action for leukemogenic effects. Therefore, the strength of the evidence for formaldehyde as a potential causal agent for human myeloid leukemia neither supports a "known" classification, nor rises to the level of "reasonably anticipated", based on NTP's classification requirements described in the Background Document for Formaldehyde (NTP 2010a).

We continue to recommend that the NTP staff and the Board of Scientific Counselors apply a strength-of-evidence approach in separately evaluating all human, animal and mechanistic data relevant to each of the three identified cancer types ― i.e. nasopharyngeal carcinoma, sinonasal adenocarcinoma, and myeloid leukemia. The scientific evidence for these three cancers varies widely and warrants independent reviews and findings. Moreover, although the NTP may generally classify formaldehyde based upon a single tumor type, we recommend that the NTP both apply the RoC classification criteria to each separate tumor type, and justify the listing classification with respect to each such endpoint. Thus, even if NTP were to conclude that formaldehyde is a known human carcinogen with respect to one (but not a second or third) endpoint, the NTP findings should clearly delineate both the scientific bases for, and the NTP RoC classification concerning, each separate cancer type. This is particularly important because, as our comments clearly delineate, the strength of the evidence for formaldehyde does not rise to the level necessary to qualify, according to the RoC classification scheme, formaldehyde as either a known or reasonably anticipated human myeloid leukemogen when a comprehensive strength-of-evidence evaluation is performed on the full range of existing scientific data.

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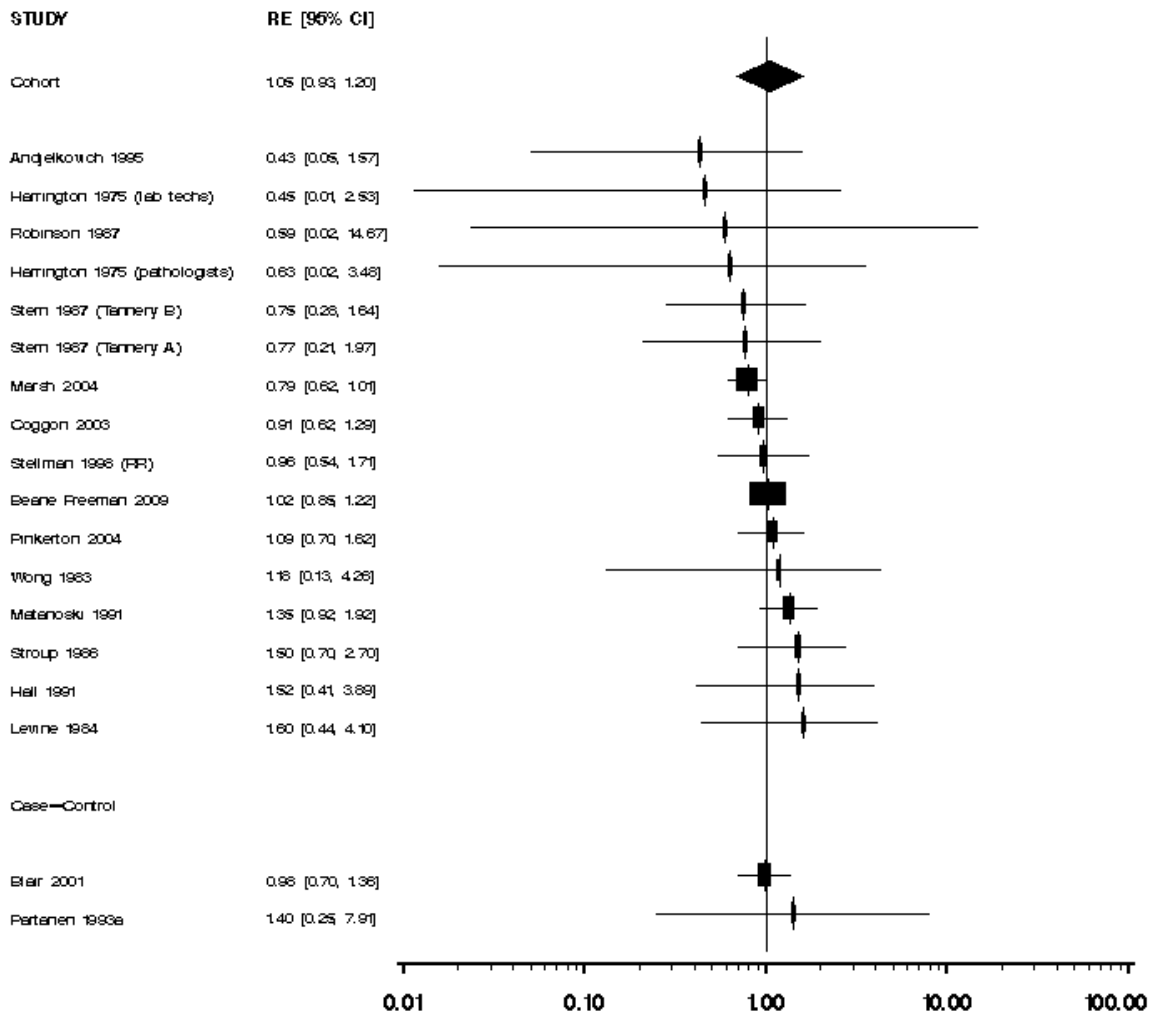

Figure 1: Forest plot by study design for leukemia

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